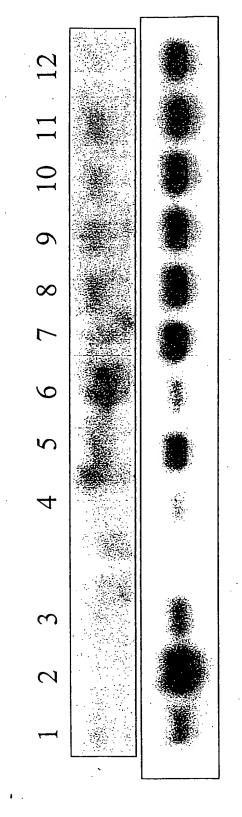
mRNA Reverse Transcriptase Primers: YT.....<sub>15</sub>U<sub>2</sub>AG<sub>a</sub>  $G_7AU_2dT$ : cDNA 7 (cDNA normalization by PCR with Primers: GAPDH  $G_7AU_2T$ :  $G_7TATTTATTTAT$  $T_{15}U_2AG_n$ Final product 6% native PAAG Fragment elution, PCR, cloning and verification of differential expression Sequence analysis, cDNA cloning





5. Small Intestine 6. Ovary 2. Skeletal muscle

1. Colon

7. Brain

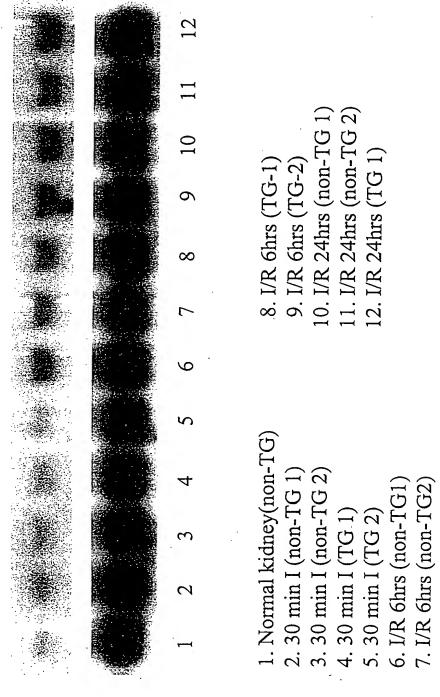
8-12. Kidney samples

4. Spleen

3. Liver

Lower panel: the same blot hybridized with mouse GAPDH gene probe, shown as Upper panel: Northern blot hybridized with candidate gene probe internal control

Sheet No. 2 of 7 Express Mail No. EL740476596US
Docket No. 266/171
Shorth Figure



Sheet No. 3 of 7 Docket No. 266/171 Express Mail No. EL740476596US

Docket No. 266/171 Sheet No. 4 of 7

Express Mail No. EL740476596US

1. Molecular marker

. Total cell extract

. 20,000g ppt after sonication

4. 180,000g ppt

5. 20%  $(NH_4)_2SO_4$ 

6.  $40\% (NH_4)_2 SO_4 p$ 

**↑** IAP

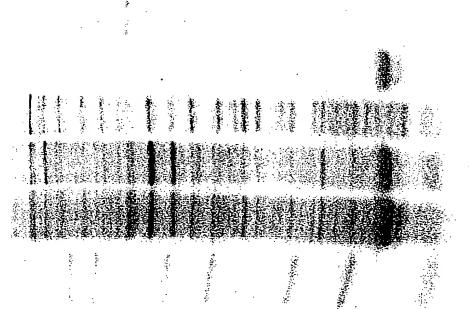
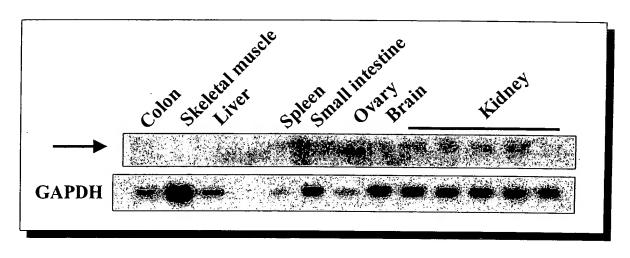


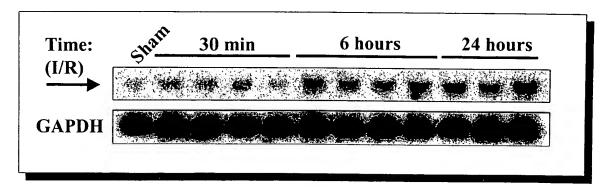
Figure 5

Express Mail No. EL740476596US Docket No. 266/171 Sheet No. 5 of 7

## mRNA expression in mouse tissues

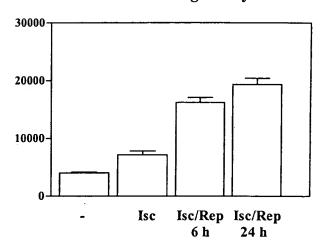


## mRNA expression after kidney ischemia/reperfusion



Express Mail No. EL740476596US Docket No. 266/171 Sheet No. 6 of 7

## Measurement of IAP mRNA activation during kidney I/R \*

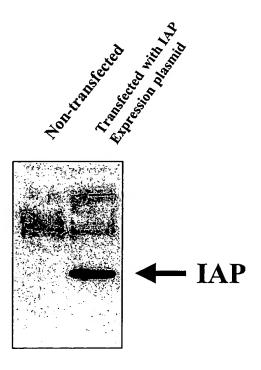


\*At least 4 animals were analyzed per each group.

Data are expressed as the mean +SEM.

Figure 6

## Overexpression of IAP in Hela cells



Hela cells were transfected with plasmid DNA, containing pEF/myc/cyto vector (Invitrogen) containing human IAP cDNA. After 48 hrs cells were lysed and total cellular extract was analyzed by Western blotting using anti-IAP rabbit polyclonal Ab and Phototope-HRP Western Blot Detection Kit (BioLabs) as described in manufacturer's protocol.